# HELLER EHRMAN WHITE & MCAULIFFE LLP Sheet 1 of 8 Title METHODS FOR THE PRODUCTION OF REDOX PROTEINS. Applicant van Rooijen et al. Serial No. 10/032,201 Filed: December 19, 2001 Our Docket No.: 38814-351B

10 20	30	40 50 60
TR A T G A A T G G T C T C G A A A C T C A ATTHIREDB A T G A A T G G T C T C G A A A C T C A	C A A C A C A A G G C T C T G T A C A A C A C A A G G C T C T G T A	T C G T A G G A A G T G G C C C A G C G G C A T C G T A G G A A G T G G C C C A G C G G C A
70 80	90	100 110 120
TR CACACGGCGGCGATTTACGCATTHIREDBCACACGGCGGCGATTTACGC	AGCTAGGGCTGAACTTA	A A C C D C D D C D C D D C C A A C C A D C C
130 140		
TR ATGGCTAACGACATCGCTCC	CGGTGGTCAACTLAACAA	160 170 180 C C A C C G A C G T C G A G A A T T T C
ATTHIREDB A T G G C T A A C G A C A T C G C T C C		C C A A C C A C C G C G T - G A G A A T T T C
TR CCCGGATTTCCAGAAGGTAT	TCTCGGAGTAGAGCTCA	220 230 240 C T G A C A A A T T C C G T A A A C A A T C G
ATTHIREDE C C C G G A T T T C C A G A A G G T A T	TCTCGGAGTAGAGCTCA	CTGACAAATTCCGTAAACAATCG
TR GAGCGATTCGGTACTACGAT	270	280 290 300 C G A A A G T C G A T T T C T C T T C G A A A
ATTHIREDB G A G C G A T T C G G T A C T A C G A T	ATTTACAGAGACGGTGA	CGAAAGTCGATTTCTCTTTCGAAA
310 320 TR CCGTTTAAGCTATTCACACA	330	340 350 360
ATTHIREDB C C G T T T A A G C T A T T C A C A G A	TTCAAAAGCCATTCTCG TTCAAAAGCCATTCTCG	C T G A C G C T G T G A T T C T C G C T A C T C T G A C G C T G T G A T T C T C G C T A T C
370 380	390	400 420
TR GGAGCTGTGGCTAAGCGGCT ATTHIREDBGGAGCTGTGGCTAAGTGGCT	TAGCTTCGTTGGATCTG	GTGAAGGTTCTGGAGGTTTCTCC
430 440	450	
TR AACCGTGGAATCTCCGCTTG	TGCTGTTTGCGACGGAC	460 470 480 C T G C T C C G A T A T T C C G T A A C A A A
ATTHIREDB A A C C G T G G A A T C T C C G C T T G		
TR CCTCTTGCGGTGATCGGTGG	510 AGGCGATTCAGCAATGG	520 530 540 A A G A A G C A A A C T T T C T T A C A A A A
ATTHIREDB CCTCTTGCGGTGATCGGTGG	AGGCGATTCTGCAATGG	A A G A A G C A A A C T T T C T T A C A A A A
550 560 TR TATGGATCTAAAGTGTATAT	AATCCATAGGAGAGATG	580 590 600 C T T T T A G A G C G T C T A A G A T T A T G
ATTHIREDB TATGGATCTAAAGTGTATAT	AATCGATAGGAGATG	CTTTTAGAGCGTCTAAGATTATG
610 620 TR CAGCAGCGAGCTTTCTCTCTA	630	640 650 660
ATTHIREDS CAGCAGCTTTGTCTAA	T C C T A A G A T T G A T G T G A T C C T A A G A T T G A T G T G A	TTTGGAACTCGTCTGTTGTGGAA
670 680	600	700
TR GCTTATGGAGATGGAGAAG ATTHIREDB GCTTATGGAGAAAG	A G A T G T G C T T G G A G G A T A G A T G T G C T T G G A G G A T	M C
730 740	750	760
TR GGAGATGTTTCTGATTTAAA ATTHIREDBGGAGATGTTTCTGATTAAA	AGTTTCTGGATTGTTCT	**************************************
TR ACCAAGTTTTTGGATGGTGG	810 T G.T T G A G T T A G A T T C G G	820 830 840 A T G G T T A T G T T G T C A C G A A G C C T
THE THE CARGITTTTTGGATGGTGG	TGTTGAGTTAGATTCGG	ATGGTTATGTTGACGAAGCCT
TR GGTACTACACAGACTAGCGT	870 T C C C G G A G T T T T C G C T G	880 890 900 C G G G T G A T G T T C A G G A T A A G A A G
ATTHIREDS G G T A C T A C A C A G A C T A G C G T	TCCCGGAGTTTTCGCTG	CGGGTGATGTTCAGGATAAGAAG
TR TATAGGCAAGCCATCACTGC	930	940 950 960 T G G C A G C T T T G G A T G C A G A G C A T
ATTHIREDB TATAGGCAAGCCATCACTGC	TGCAGGAACTGGGTGCA	T G G C A G C T T T G G A T G C A G A G C A T T G G C A G C T T T G G A T G C A G A G C A T
970 980 TR TACTTACAAGAGATTGGATG	990	1000 1010 1020
TR TACTTACAAGAGATTGGATCATTHIREDBTACTTACAAGAGATTGGATC	T C A G C A A G G T A A G A G T G T C A G C A A G G T A A G A G T G	ATTGA ATTGA
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FIG. I

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Translation of ATTHIREDB M N G L E T H N T R L C Translation of TR M N G L E T H N T R L C	LCIVGSGP	20 A A H T A A I Y A A A H T A A I Y A	30 ARAELKPLLF ARAELKPLLF	40 EGWMANDIA EGWMANDIA	50 PGGQLNQPP-RE PGGQLTTTTDVE	N F
Translation of ATTHIREDB PGFPEGILG Translation of TR	70 LGVELTDKFRK C LGVELTDKFRK C	80 QSERFGTTI QSERFGTTI	90 FTETVTKVDF FTETVTKVDF	SSKPFKLFT SSKPFKLFT	110 DSKAILADAVIL DSKAILADAVIL	120 A T
Translation of ATTHIREDB G A V A K W L S F V G Translation of TR	S G E V L G G	140 L W N R G I S A C F W N R G I S A C	150 A V C D G A A P I F A V C D G A A P I F	160 RNKPLAVIG RNKPLAVIG	170 G G D S A M E E A N F L G G D S A M E E A N F L	180 T K
Translation of ATTHIREDB Y G S K V Y I I D R R D Y G S K V Y I I H R R D Y G S K V Y I I H R R D	A F R A S K A F R A S K	1 M Q Q R A L S N I M Q Q R A L S N	210 PKIDVIWNSS PKIDVIWNSS	220 V Y E A Y G D G E V Y E A Y G D G E	230 R D V L G G L K V K N V R D V L G G L K V K N V	240 V T
Translation of ATTHIREDB G D V S D L K V S G L Translation of TR	FFAIGHE	260 PATKFLDGG PATKFLDGG	270 VELDSDGYVV VELDSDGYVV	280 T K P G T T Q T S T K P G T T Q T S	290 V P G V F A A G D V Q D V P G V F A A G D V Q D	X K K
Translation of ATTHIREDB Y R Q A I T A A G T G C Translation of TR	GCMAALDA GCMAALDA	320 EHYLQEIGS EHYLQEIGS	330 Q Q G K S D Q Q G K S D	340	350	360

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y 22 March Mars

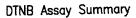
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	700 $708$ $708$ $708$ $708$ $708$ $708$ $708$ $708$ $708$ $708$ $709$ $708$ $709$ 709			$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	$320 \qquad 330 \qquad 330 \qquad 340 \qquad 350$ $350 \qquad 20 \  \                             $	380 $1360$ $1360$ $1370$ $1380$ $1$	$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$
M.lep TR/Trxh M N $\frac{T}{G}$ Arab TR-link-Trxh M N $\frac{T}{G}$	M.lep TR/Trxh Arab TR-link-Trxh	M.lep TR/Trxh $\begin{array}{c} \mathbb{C} & \mathbb{A} & \mathbb{T} \\ Arab & TR-link-Trxh & \mathbb{C} & \mathbb{A} & \mathbb{V} \end{array}$	M.lep TR/Trxh S K I Arab TR-link-Trxh S K I	M.lep TR/Trxh V V Arab TR-link-Trxh K V	M.lep TR/Trxh $\frac{\text{V}}{\text{D}}$ Arab TR-link-Trxh $\overline{\text{Q}}$ D	M.lep TR/Trxh Arab TR-link-Trxh D w	M.lep TR/Trxh Arab TR-link-Trxh C G	M.lep TR/Trxh $\frac{\mathbb{Q}}{\mathbb{R}} \frac{\mathbb{G}}{\mathbb{E}}$

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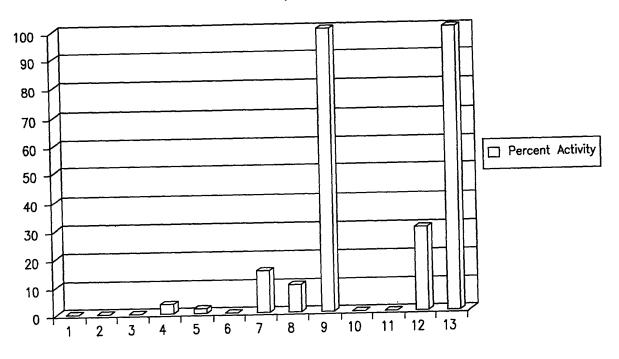


FIG. 4

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## **HETEROMULTIMERS**

Class Heteromultimer		Example sequence reference for		
		heteromultimeric subunits		
Biosynthetic	3-methyl-2-oxobutanoate	McKean, et al. Biochim. Biophys. Acta (1992)		
	dehydrogenase (2-oxoisovalerate	1171:109-112 / Chuang, J.L., et al FEBS Lett. a		
	dehydrogenase (lipoamide))- E1	(1990) 262 (2), 305-309.		
	component)	, , , , , , , , , , , , , , , , , , , ,		
Biosynthetic	3-oxoadipate CoA-transferase	Parales, R.E. and Harwood, S.C. J. Bacteriol. (1992)		
		174:4657-4666		
Biosynthetic	anthranilate synthase:indole-3-glycerol	Zalkin, H.; et al. J. Biol. Chem. (1984) 259:3985-		
	phosphate synthase	3992.		
Biosynthetic	beta-ketoacyl-[acyl carrier protein]	Siggaard-Andersen, M. et al. Proc. Natl. Acad. Sci.		
	synthase I	U.S.A. (1991) 88:4114-4118		
Biosynthetic	butyrateacetoacetate CoA-transferase	Fischer, R.J., et al. J. Bacteriol. (1993) 175 (21),		
		6959-6969.		
Biosynthetic	cAMP dependent protein kinase	Mutzel, R et al. Proc. Natl. Acad. Sci. U.S.A. (1987)		
		84:6-10./ Burki, E., et al. Gene (1991) 102 (1), 57-		
		65.		
Biosynthetic	carbamoyl-phosphate synthase	Shigenobu, S., et al. Nature. (2000) 407 (6800), 81-		
		86.		
Biosynthetic	Creatine kinase	Billadello, J.J.; et al. Biochem. Biophys. Res.		
		Commun. (1986) 138:392-398. / Roman, D.; et al.		
		Proc. Natl. Acad. Sci. U.S.A. (1985) 82:8394-8398.		
Biosynthetic	gamma-glutamyltransferase (gamma-	Papandrikopoulou, A.; et al. Eur. J. Biochem.		
	glutamyl transpeptidase)	(1989) 183:693-698.		
Biosynthetic	glutathione transferase	Morrow, C.S. et al. Gene (1989) 75:3-11		
Biosynthetic	glycerol-3-phosphate dehydrogenase	Cole, S.T. et al. J. Bacteriol. (1988) 170:2448-2456.		
Biosynthetic	guanylate cyclase	Hinsch, K.D. et al. FEBS Lett. (1988) 239:29-34/		
		Koesling, D. et al. FEBS Lett. (1990) 266:128-132.		
Biosynthetic	heterodisulfide reductase	Smith, D.R., et al. J. Bacteriol. (1997) 179 (22),		
		7135-7155.		
Biosynthetic	human cathepsin	Ritonja, A. et al. FEBS Lett. (1988) 228:341-345.		
Biosynthetic	Hydrogenase	Menon, N.K. et al. J. Bacteriol. (1990) 172:1969-		
		1977.		
Biosynthetic	Meprin A	Johnson, G.D. and Hersh, L.B. J. Biol. Chem.		
	•	(1992) 267:13505-13512.		
Biosynthetic	methionine adenosyltransferase	Horikawa, S.; Tsukada, K. FEBS Lett. (1992)		
	•	312:37-41.		
Biosynthetic	methylmalonyl-CoA mutase	Jackson, C.A. et al. Gene (1995) 167:127-132.		
Biosynthetic	mitochondrial processing peptidase	Pollock, R.A. et al. EMBO J. (1988) 7:3493-3500.		
Biosynthetic	Na+/K+-exchanging ATPase	Shull, G.E., et al. Biochemistry (1986) 25 (25),		
		8125-8132./Mercer,R.W., et al.		
		Mol. Cell. Biol. (1986) 6 (11), 3884-3890./		
		Mercer, R.W., et al. J. Cell Biol. (1993) 121 (3),		
		579-586.		
Biosynthetic	NAD(+)-dependent isocitrate	Cupp, J.R. and McAlister-Henn, L. J. Biol. Chem.		
	dehydrogenase	(1992) 267:16417-16423. /Cupp, J.R. and		
		McAlister-Henn, L.		
		J. Biol. Chem. (1991) 266:22199-22205.		
Biosynthetic	phosphoribosylformylglycinamidine	Ebbole, D.J.; Zalkin, H. J. Biol. Chem. (1987)		
- -	synthase	262:8274-8287.		
Biosynthetic	protocatechuate 3,4-dioxygenase	Frazee, R.W.; et al. J. Bacteriol. (1993) 175:6194-		
-	. along gonuse	6202.		
Biosynthetic	S-100 protein	Engelkamp, D.; et al. Biochemistry (1992)		
		Linguistanip, D., et al. Diochemistry (1992)		

FIG. 5A

Electron transport

Electron transport

cytochrome-c3 hydrogenase

electron transfer flavoprotein

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		31:10258-10264. / Allore, R.J.; et al. J. Biol. Chem. (1990) 265:15537-15543.
Biosynthetic	sucrosefructan 6-fructosyltransferase	Sprenger, N.; et al. Proc. Natl. Acad. Sci. U.S.A. (1995) 92:11652-11656.
Biosynthetic	Superoxide dismutase	Capo, C.R.; et al. Biochem. Biophys. Res. Commun. (1990) 173:1186-1193.
Biosynthetic	Urease	Labigne, A.; et al. J. Bacteriol. (1991) 173:1920-1931.
Biosynthetic	urokinase-type plasminogen activator (urokinase)	Belin, D. et al. Eur. J. Biochem. (1985) 148:225-232.
Biosythetic	methylmalonyl-coenzyme A mutase	Birch, A., et al J. Bacteriol. (1993) 175 (11), 3511-3519.
Calcium binding	Calcineurin	Muramatsu, T. and Kincaid, R.L. Biochim. Biophys. Acta (1993) 1178 (1), 117-120 / Guerini, D. et al. DNA (1989) 8:675-682.
Calcium binding	Calgranulin	Imamichi, T. et al. Biochem. Biophys. Res. Commun. (1993) 194:819-825.
Calcium binding	Calpain	Aoki, K. et al. FEBS Lett. (1986) 205:313-317.
DNA binding	API	van Straaten, F., et al. Proceedings of the National Academy of Sciences of the United States of America. (1983) 80 (11), 3183-3187. /Hattori, K., et al Proceedings of the National Academy of Sciences of the United States of America. (1988) 85 (23), 9148-9152.
DNA binding	сМус-Мах	Schreiber-Agus, N et al. Mol. Cell. Biol. (1993) 13 (5), 2765-2775.
DNA binding	DNA binding protein HU-1/HU-2	Laine, B. et al. Eur. J. Biochem. (1980) 103:447-461.
DNA binding	hepatic nuclear factor 1	Bach, I. et al. Nucleic Acids Res. (1992) 20 (16), 4199-4204. / Rey-Campos, J. et al. EMBO J. (1991) 10 (6), 1445-1457.
DNA binding	Integration host factor	Miller, H.I. Cold Spring Harbor symposia on quantitative biology. (1984) 49, 691-698. / Flamm, E. and Weisberg, R.A. J. Mol. Biol. (1985) 183:117-128.
DNA binding	Ku	Reeves, W.H. and Sthoeger, Z.M. J. Biol. Chem. (1989) 264 (9), 5047-5052. / J. Biol. Chem. (1989) 264 (23), 13407-13411.
DNA binding	MutS	Bocker et al. 1999. Cancer Research 59, 816-822.
DNA binding	NF-E2	Chan, J. Y. et al. Proc. Natl. Acad. Sci. U.S.A. (1993) 90 (23), 11366-11370./ Toki, T., et al. Oncogene (1997) 14 (16), 1901-1910.
DNA binding	nuclear factor kB (NFkB)	Kieran M, et al. Cell. (1990) Sep 7;62(5):1007-18./ Ruben SM, et al. Science (1991) Mar 22;251(5000):1490-3. Erratum in: Science (1991) Oct 4;254(5028):11
Electron transport	corrinoid/iron-sulfur protein	Lu, W.P. et al. J. Biol. Chem. (1993) 268:5605- 5614.
Electron transport	cytochrome d ubiquinol oxidase	Green, G.N. et al. J. Biol. Chem. (1988) 263:13138-

FIG. 5B

13143.

Menon, N.K. et al. J. Bacteriol. (1987) 169:5401-

Finocchiaro, G. et al. Biol. Chem. (1988) 263:15773-15780. / Finocchiaro, G. et al. Eur. J.

Biochem. (1993) 213:1003-1008.

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Electron transport	xylene monooxygenase	Shaw I P and Harayama S Em I Dischar
	Aylone monooxygenase	Shaw, J.P. and Harayama, S. Eur. J. Biochem. (1992) 209:51-61. / Kasai, Y., et al. J. Bacteriol.
Growth factor	hepatocyte growth factor	(2001) 183 (22), 6662-6666.
Growth factor	human chorionic gonadotropin	Nakamura, T. et al. Nature (1989) 342:440-443.
	•	Morgan, F.J. et al. J. Biol. Chem. (1975) 250 (13), 5247-5258.
Growth factor	Platelet-derived growth factor	Takimoto, Y., et al. Hiroshima J. Med. Sci. (1993) 42 (1), 47-52./ Josephs, S.F., et al. Science (1984) 225 (4662), 636-639.
Hormone	Bombyxin	Adachi, T. et al. J. Biol. Chem. (1989) 264:7681-7685.
Hormone	Follicle stimulating hormone	Fiddes, J.C. and Goodman, H.M. J. Mol. Appl. Genet. (1981) 1 (1), 3-18. / Watkins, P.C., et al. DNA (1987) 6 (3), 205-212.
Hormone	Insulin	Bell,G.I., Pictet,R.L., Rutter,W.J., Cordell,B., Tischer,E. and Goodman,H.M. Sequence of the human insulin gene. Nature. 284 (5751), 26-32 (1980)
Hormone	Luteinizing Hormone	Fiddes, J.C. and Goodman, H.M. J. Mol. Appl. Genet. (1981) 1 (1), 3-18. / Shome, B. and Parlow, A.F. J. Clin. Endocrinol. Metab. (1973) 36 (3), 618-621.
Hormone	Thyroid stimulating hormone	Fiddes, J.C. and Goodman, H.M. J. Mol. Appl. Genet. (1981) 1 (1), 3-18. / Hayashizaki Y, et al. FEBS Lett. (1985) 188 (2), 394-400.
Immune	B-cell antigen receptor complex	Hashimoto, S. et al. J. Immunol. (1993) 150 (2), 491-498. / Flaswinkel, H. and Reth, M. Immunogenetics (1992) 36 (4), 266-269.
Immune	Cell surface CD8 molecules	Ureta-Vidal, A., et al. Immunogenetics (1999) 49 (7-8), 718-721.
Immune	human complement subcomponent Clq	Sellar, G.C. et al. Biochem. J. (1991) 274:481-490.
Immune	T cell receptor	Talken,B.L. et al. Scand. J. Immunol. (2001) 54 (1-2), 204-210.
Photosynthesis	C-phycocyanin	Offner, G.D. et al. J. Biol. Chem. (1981) 256:12167-12175. / Troxler, R.F. et al. J. Biol. Chem. (1981) 256:12176-12184.
Photosynthesis	ferroredoxin-thioredoxin reductase	Chow, L.P. et al. Eur. J. Biochem. (1995) 231:149- 156. / Iwadate, H. et al. Eur. J. Biochem. (1994) 223:465-471.
Photosynthesis	Light harvesting complex I	Proc. Natl. Acad. Sci. U.S.A. (1984) 81, 189-192.
Photosynthetic	cytochrome b559	Carrillo, N. et al. Curr Genet. 1986;10(8):619-24.
Protease	ATP-dependent Clp protease	Gerth, U. et al. Gene (1996) 181:77-83. / Kunst,F. et al. Nature (1997) 390 (6657), 249-256.
Receptor	alpha-2-macroglobulin receptor	Strickland, D.K. et al. J. Biol. Chem. (1990) 265:17401-17404. / Strickland, D.K. et al. J. Biol. Chem. (1991) 266:13364-13369.
Receptor	Interleukin-2 receptor	Ishida, N. et al. Nucleic Acids Res. (1985) 13:7579-7589. / Hatakeyama, M. et al. Science (1989) 244:551-556 / Takeshita, T. et al. Science (1992) 257:379-382.
Receptor	platelet-derived growth factor receptor	Lee, K.H. et al. Mol. Cell. Biol. (1990) 10:2237- 2246. / Herren, B. et al. Biochim. Biophys. Acta 1173 (3), 294-302 (1993).
Structural	Hemoglobin	Heindell, H.C. et al. Cell (1978) 15 (1), 43-54.

FIG. 5C

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		Best, J.S. et al. Hoppe-Seyler's Z. Physiol. Chem. (1989) 350 (5), 563-580. / Hardison, R.C. J. Biol. Chem. (1981) 256 (22), 11780-11786.
Structural	human platelet glycoprotein Ib	Wenger,R.H. et al. Biochem. Biophys. Res. Commun. (1988) 156 (1), 389-395. / Yagi,M. et al. J. Biol. Chem. (1994) 269 (26), 17424-17427.
Structural	Plasma fibronectin	Kornblihtt, A.R. et al. Proc. Natl. Acad. Sci. U.S.A. (1983) 80:3218-3222.
Structural	Spectrin	Sahr, K.E. et al. J. Biol. Chem. (1990) 265:4434-4443. / Winkelmann, J.C. et al. J. Biol. Chem. (1990) 265:11827-11832.
Structural	Tubulin	Ponstingl, H. et al. Proc. Natl. Acad. Sci. U.S.A. (1981) 78:2757-2761. / Krauhs, E. et al. Proc. Natl. Acad. Sci. U.S.A. (1981) 78:4156-4160.
Toxin	Agkisacutacin	Cheng, X. et al. Biochem. Biophys. Res. Commun. (1999) 265 (2), 530-535.
Toxin	Beta bungarotoxins	Kondo, K. et al. J. Biochem. (1978) 83:101-115.
Toxin	Crotoxin	Bouchier, C. et al. Nucleic Acids Res. (1988) 16 (18), 9050.
Toxin	Mojave toxin	John, T.R. et al. Gene (1994) 139:229-234.
Toxin	venom protein C9S3	Rowan, E.G. et al. Nucleic Acids Res. (1990) 18:1639. / Joubert, F.J. and Viljoen, C.C. Hoppe- Seyler's Z. Physiol. Chem. (1979) 360:1075-1090.
Miscellaneous	Inhibin	Forage, R.G. et al. Proc. Natl. Acad. Sci. U.S.A. (1986) 83:3091-3095.
Miscellaneous	Monellin	Frank, G. and Zuber, H. Hoppe-Seyler's Z. Physiol. Chem. (1976) 357:585-592.
Miscellaneous	mRNA capping enzyme	Niles, E.G. et al., J. Virology (1986) 153:96-112.
Miscellaneous	Soybean insulin-binding protein si30	Barbashov, S.F. et al. Bioorg. Khim. (1991) 17:421-423.

FIG. 5D